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Immune complex systems in the nephritis of systemic lupus erythematosus

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At present there is considerable evidence that the pathologic processes in the nephritis of systemic lupus erythematosus (SLE) are mediated at least in part by immunological mechanisms [1–7]. The deposition of immune complexes in the glomeruli in this disease and the subsequent injury that ensues bear a resemblance to the nephritis in the experimental serum sickness animal models. The immunological and phlogistic events following the injection of a single foreign antigen have been defined to a considerable degree in these models. However, it is well known that marked variations in the frequency of induction and severity of the nephritis occur in different animals because of individual variations in the immune response [8, 9]. The situation in human systemic lupus erythematosus is considerably more complex since in man the immune response is certainly no less variable, and it now appears that several antigen-antibody systems are involved in the pathogenesis of the renal lesion.

It is the purpose of this paper to review some of the serologic and renal biopsy studies that have helped to delineate the heterogeneity of the immune complex systems that appear to be important.

Immunofluorescent studies of renal biopsies. Morphologic data obtained from immunofluorescence studies of renal biopsies from a broad group of SLE patients suggest that different immune complexes are involved in the nephritic process (Table 1). Marked variations in the deposition of γ-globulin and complement can occur as indicated by the presence of four distinct immunofluorescence patterns of glomerular staining (Fig. 1). Linear staining appeared as homogenous staining of the glomerular basement membrane. Mesangial patterns were characterized by deposits lying between loops that appeared to radiate from the central portion of the glomerulus. Granular deposits ranged in size and were distributed irregularly throughout the glomerulus. Lumpy deposits of γ-globulin outlined the glomerular tufts obscuring the normal architecture. Linear, granular or lumpy types of deposits were usually accompanied by mesangial deposits of immunoglobulin and complement. Immunoglobulin and complement were observed in similar distribution except in the case of the linear staining pattern which was confined to γG-globulin. An occasional renal biopsy with granular deposits showed very few deposits in the mesangium.

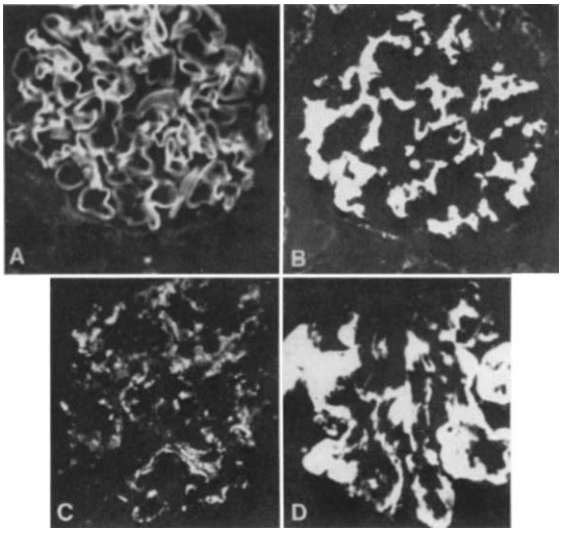

Fig. 1. *Different patterns of fluorescence in four SLE biopsies. A)* Patient D. S.: linear deposits of γG-globulin along GBM. *B)* Patient P. W.: mesangial deposits of γ-globulin shown as irregular strandlike areas of fluorescence lying between capillary loops. *C)* Patient J. B.: granular deposits of γ-globulin along GBM. *D)* Patient M. R.: lumpy deposits of immunoglobulin outlining tufts. All × 250.

Certain patterns of glomerular deposits appear to correlate with the presence of renal disease. Linear or mesangial staining alone were found in kidneys without clinical or

Table 1. Clinical findings and renal biopsy immunofluorescent data in SLE patients

Patient	Renal biopsies: Pattern of deposits	Protein deposited[a]: γG	γM	β_1C	Proteinuria[b] *g/24 hr*	Serum creatinine *mg/100 ml*[c]	Duration (years): Total disease	Renal disease	Prednisone: *mg/day*	Duration *years*[d]
1. MCK	Membranous	2+	0	0	0.10	0.9	10	—	2.5	7
2. SCH	Membranous	3+	0	0	0.20	0.9	3	—	50 → 2.5	1
3. BER	Mesangial	±	0	1+	0	0.9	3	—	60 → 10	1
4. GAM	Mesangial	2+	±	2+	0	1.0	7	—	0	0
5. URA	Mesangial	3+	0	3+	0.1	1.1	4	—	60 → 5	1
6. EGL	Mesangial	2+	±	2+	0.1	0.7	2	—	120 → 20	0.25
7. BAN	Mesangial	3+	1+	3+	0.31	1.0	6	—	0	0
8. WOL	Mesangial	2+	±	2+	0.33	0.8	0.06	—	40	0.04
9. CIA	Granular	2+	2+	2+	0.8	1.2	12	12	0[e]	0
10. ROS	Granular	2+	±	2+	1.5	1.1	2	1	20 → 2.5	0.25
11. HAU	Granular	3+	3+	3+	1.8	1.1	29	2	0	0
12. LOO	Granular	2+	0	2+	2.2	0.8	1	0.5	60	0.06
13. WIL	Granular	3+	1+	3+	2.9	1.2	1	1	80 → 35	1
14. HAR	Granular	3+	2+	2+	3.1	1.2	9	0.17	5 → 10	6
15. FOR	Granular	3+	0	3+	4.0	1.1	7	2	40 → 75	2
16. SAC	Granular	3+	1+	3+	4.4	1.4	1	0.25	40 → 15	0.25
17. MOR	Granular	2+	2+	3+	5.0	1.3	5	1	0	0
18. RUS	Lumpy	1+	3+	3+	7.6	1.6	12	2	60 → 100	0.02
19. WEI	Lumpy	±	3+	2+	8.3	2.3	2	2	10	1

[a] Graded as 0, neg; ±, trace; 1+, minimal; 2+, moderate; 3+ marked.
[b] Upper limit for normals 0.4.
[c] Upper limit for normals 1.5.
[d] Duration of therapy prior to biopsy.
[e] 40–10 mg seven years, none for three years prior to biopsy.

histological evidence of glomerular injury. The linear localization of γ-globulin in SLE appears analogous to the pattern of staining found in Goodpasture's syndrome [8]; however, the absence of associated complement deposition and nephrotoxicity differentiate the two situations. Granular deposits were associated with active glomerular nephritis, while the lumpy pattern occurred with more advanced histological and clinical renal disease.

Serologic studies. A summary of the wide variety of antibodies to autologous tissue and plasma antigens that occur in the serum of patients with SLE is shown in Table 2. The antinuclear groups have received special study, which initially was due primarily to their relationship to the LE phenomenon and their striking appearance in fluorescence antibody studies.

Antibodies to deoxyribonucleic acid were among the earliest described [10, 2]. Subsequently antibodies to both native (NDNA) and single-stranded determinants (SDNA) have been extensively characterized [4, 11–23]. Antibodies reactive with particulate and soluble nucleoprotein differ from antiDNA antibodies with respect to the requirement of a protein-DNA complex [24, 25]. Antibodies to two soluble but distinct nuclear antigens have been described. These have been partially characterized and are the Sm antigen which is periodate-sensitive [26] and ENA which is sensitive in part to both RNAse and trypsin [27, 28]. Antibodies reactive with double-stranded RNA (DSRNA) have also been demonstrated and have received considerable attention recently [29–31]. Antiribosomal antibodies have been observed in association with renal disease [32], and a variety of other anticytoplasmic antibodies have been found in SLE sera in relatively low incidence [33, 34].

Recent extensive studies on the incidence of antibodies to polynucleotides in human sera in different conditions [18]

Table 2. Antibodies in SLE sera

I. Antibodies to Nuclear Constituents
  a. Native DNA
  b. Single strand DNA
  c. Nucleoprotein
  d. "Carbohydrate-Protein" Antigens
    Sm Antigen and Others
  e. RNA-Protein (ENA)

II. Antibodies to Cytoplasmic Constituents
  a. Ribosomes (RNAase sens.)
  b. Carbohydrate Antigens
  c. Lipid Antigens

III. Antibodies to Clotting Factors

IV. Red Cell Antibodies

V. Platelet Antibodies

VI. RNA Antibodies
  Poly A · Poly U
  Poly I · Poly C
  Poly A

have indicated that significant titers of antibodies to native DNA are almost exclusively restricted to patients with SLE, whereas antibodies to DSRNA preparations (Poly A · Poly U, Poly I · Poly C) are sometimes found in sera from patients with other diseases (Table 3). Antibodies to SDNA occur more frequently than antibodies to NDNA in patients with SLE but are also present in high incidence in other disease states. Antibodies to ribonucleoproteins were limited mainly to patients with SLE, rheumatoid arthritis and mixed connective tissue disease.

Serial studies where antibody titers have been followed during disease exacerbations have proven particularly informative. An example is shown in Fig. 2. Antibody titers to NDNA, SDNA, DSRNA and RNA-protein were followed over 24 months. Two episodes of clinical activity occurred during this period and were associated with rises in antiNDNA antibody titers. Steroid treatment was associated with clinical remission with a concomitant decrease in antiNDNA antibody. Antibodies to SDNA were also present during the first episode and persisted after clinical symptoms abated. Antibodies to DSRNA appeared during clinical quiescent periods. Antibodies to RNA-protein were absent throughout the course. This example demonstrates some of the common findings from a similar study of 25 patients followed over a period from six months to four years. Peaks of activity of antibodies to NDNA, SDNA and DSRNA generally occurred sometime during the course of SLE in most patients, whereas antiribonucleoprotein antibodies were completely absent from one-third of the serial studies. Peaks of NDNA antibody appeared to correlate better with disease activity and serum complement depression than antibodies to SDNA or DSRNA. Antibodies to ribonucleoprotein when present showed little correlation with disease activity.

The NDNA-antiNDNA immune complex system. Antibodies to NDNA in significant titers are restricted primarily to patients with SLE and are useful from the diagnostic standpoint, whereas antibodies to other nuclear antigens studied occurred in patients with a variety of diseases. Further indirect evidence for the special significance of antibodies to NDNA arises from serial studies involving serum complement determinations. Fig. 3 shows the close inverse relationship between serum complement levels and NDNA antibody titers which is frequently observed during periods of disease activity in SLE patients [5, 35]. This correlation occurs during periods of disease activity in patients with and without renal disease as illustrated in Figs. 3 and 4. Fig. 3 shows the serial study of a patient whose episodes of clinical activity were characterized by nephritis. A renal biopsy during one of these periods showed deposits of γ-globulin and complement in a granular pattern along the glomerular basement membrane. In contrast, the patient whose serial study is shown in Fig. 4 had similar complement and DNA antibody findings but no clinical nephritis. A renal biopsy in this case showed only mesangial deposits.

The association of serum complement depression with the presence of antiNDNA antibodies in the serum provides indirect evidence for NDNA-antiNDNA complex formation. More direct evidence is provided by the demonstration of antigen in serum and in renal deposits. NDNA in high concentrations has been demonstrated in sera from SLE patients during periods of clinical exacerbation and was found to alternate with NDNA antibodies [5, 53]. By using fluorescein-labeled antibody specific for NDNA, NDNA has also been demonstrated in glomerular deposits in a granular pattern similar to that of γ-globulin and complement [6]. The NDNA was demonstrable after elu-

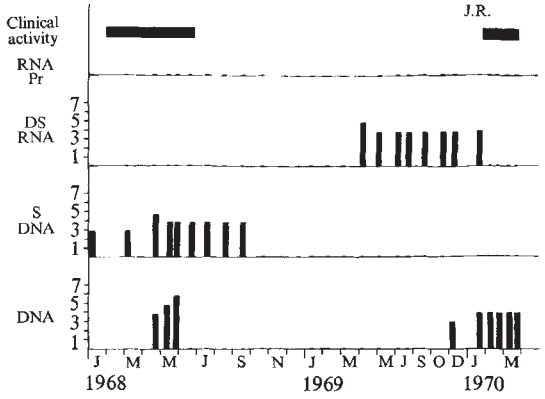

Fig. 2. *Serial study of patient J. R. showing the correlation of periods of clinical activity (horizontal bars) with various polynucleotide antibody levels in the serum.* Antibody levels (vertical bars) are given in hemagglutination titers and expressed as log base 2 on the ordinate [18].

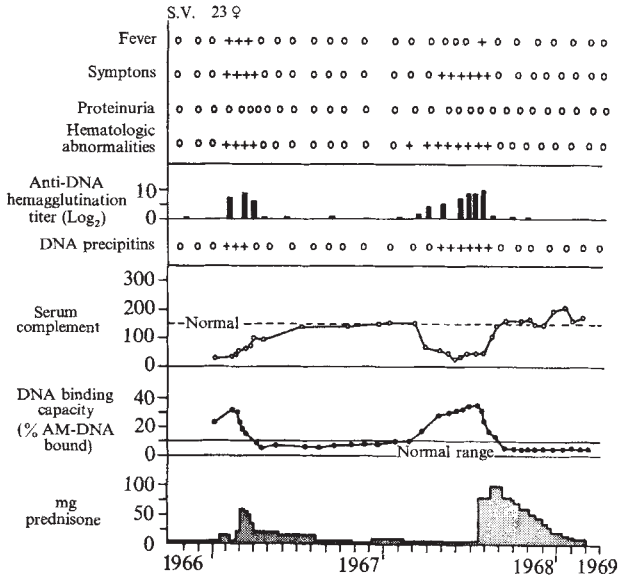

Fig. 3. *Serial study of patient S. V. showing two periods of clinical exacerbations associated with increases in titer of anti-NDNA antibodies and serum complement depression.* Antibodies were assayed by agar gel precipitation, hemagglutination and ammonium sulfate precipitation test using labeled DNA [36].

Table 3. Incidence of antibodies to polynucleotides

Sera		Antibodies positive, %				
Source	No. Tested	NDNA	SDNA	Poly A · Poly U	Poly I · Poly C	RNA Pr
SLE	60	60 (7.3)[a]	87.0 (5.1)	55.0 (5.3)	21.6 (5.0)	66.6 (12.0)
Normal	110	0.3 (5.0)[b]	3.7 (4.0)	0	1.1 (4.0)	0
Hospital[c]	65	0	16.8 (4.3)	7.7 (4.1)	9.2 (4.3)	3.1 (9.0)
Procainamide	19	0	52.6 (4.5)	10.5 (6.0)	5.3 (7.0)	0
Chronic active hepatitis	43	2.3 (3.0)	58.2 (5.1)	27.2 (6.0)[d]	—	0[d]
Infectious mononucleosis	20	0	40.0 (5.2)	10.0 (4.0)	5.0 (4.0)	0
Rheumatoid arthritis[e]	32	3.1 (3.0)	59.5 (5.5)	3.0 (5.0)	0	15.5 (10.0)
Chronic glomerulonephritis	40	2.5 (4.0)	7.5 (4.6)	0	—	2.5 (9.0)
Primary biliary cirrhosis	20	0	15.0 (3.6)	—	—	—
Carcinoma of cervix	13	0	0	0	0	

[a] Mean titer of group of serums expressed as log base 2, shown in parenthesis.
[b] Two hundred eighty normal sera were tested.
[c] Random hospital sera obtained from patients with a variety of diseases.
[d] Eleven sera were tested.
[e] Two patients had clinical evidence of mixed connective tissue disease in addition to rheumatoid arthritis.

Table 4. Concentration of antibodies in glomerular eluates [36]

	Total number studied	Anti NDNA	Anti SDNA	Anti RNA Pr[a]
Acid buffer eluates	9	5[b]	6	3
Deoxyribonuclease eluates	9	6	8	1

[a] RNA Pr, ribonucleoprotein.
[b] Number of eluates with increased antibody activity per mg of globulin in comparison with serum.

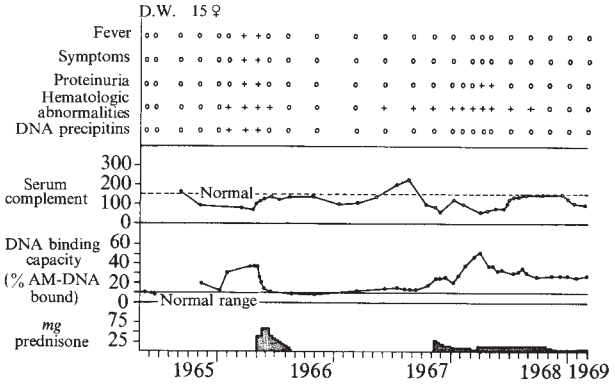

Fig. 4. *Serial study of patient D. W. showing two periods of clinical exacerbations with systemic symptoms and renal disease.* Each episode is associated with an increased titer of antiNDNA and serum complement depression [36].

tion of *in vivo* antibodies which blocked the *in vitro* reaction of fluorescein-labeled antiNDNA antibodies. AntiNDNA antibodies have been eluted from kidneys in which NDNA has been demonstrated in glomerular deposits [6].

The elution studies of isolated glomeruli from autopsy kidneys have been one of the most informative avenues of investigation in the study of SLE nephritis. The results obtained in one such study [36] are shown in Table 4. Eluates were prepared by treating glomeruli with 0.2 M, pH 2.8 glycine buffer or desoxyribonuclease. Several types of antibodies were eluted by both procedures. It was determined that certain types of antibodies were concentrated in the glomeruli if the ratio of the antibody titer to the γ-globulin content in the eluate was greater than that for a terminal serum specimen. Most eluates showed concentrations of antibodies directed against NDNA and SDNA. Antiribonucleoprotein antibodies were also concentrated in some eluates, but no antiDSRNA antibodies were found.

The SDNA-antiSDNA system. Antibodies to SDNA occur in a variety of diseases associated with active tissue destruction but the highest incidence occurs in patients with SLE. Recently SDNA antigen also has been found in the sera of a similar group of patients [37]. The highest levels of SDNA were found in patients with SLE. This was especially apparent when sera were assayed throughout the course of the disease of individual patients (Table 5). The mean serum concentration of SDNA in these serial studies was 50 μg/ml and levels as high as 250 μg/ml were observed in individual sera. SDNA alternated with the presence of SDNA antibody in a fashion similar to that demonstrated for the NDNA-antiNDNA system. In a number of sera both SDNA and antibody to SDNA could be demonstrated. In addition, evidence was presented that antiNDNA-SDNA complexes can also occur in the sera of SLE patients.

The association of rising titers of antiSDNA antibody with serum complement depression and clinical activity was less prominent than that observed for antiNDNA antibodies; however, SDNA antibodies frequently arose in parallel with antiNDNA during periods of clinical activity.

Table 5. Incidence of SDNA antigen and antibody in serial studies of SLE patients [37]

Patient	Duration of study, *months*	No. of sera tested	% of sera positive for SDNA antigen	Mean conc. of SDNA antigen (*μg/ml*) in sera	% of sera positive for SDNA antibody
Wol	24	22	0	0	23
Loo	6	11	0	0	46
Esp	25	15	0	0	54
Gam	9	21	0	0	66
Har	22	24	0	0	42
Vra	36	45	13	41.3	62
Bro	31	26	15	17.1	39
Hau	51	55	15	20.1	0
Ros	24	25	20	19.9	32
McK	16	19	21	15.2	37
For	24	24	25	19.1	54
Egl	31	34	28	22.2	21
Ber	16	28	29	18.2	32
Pea	17	24	33	25.9	83
Rus	31	40	48	65.4	32
Sil	13	15	60	111.8	27
Wil	34	22	64	125.4	27
Wei	10	20	90	52.6	20

More direct evidence that the SDNA-antiSDNA system may play a role in the pathogenesis of nephritis in SLE is the detection of both antigen and antibody in glomerular deposits in diseased kidneys. Immunofluorescence studies by Andres et al [38] have demonstrated that SDNA antigen is deposited in glomeruli lesions of SLE kidneys in association with γ-globulin and complement deposits, while elution studies (Table 3) indicate that SDNA antibody is selectively concentrated in glomeruli obtained from SLE kidneys at autopsy.

Detection of immune complexes with C1q. The C1q component of complement which is known to precipitate soluble immune complexes [39] has recently been shown to give precipitin reactions with aggregated γ-globulin or immune complexes in gel diffusion [40]. This method was found useful in detecting high molecular weight complexes containing γ-globulins in hypocomplementemic sera from SLE patients with active disease as shown in Fig. 5 [40, 41]. A number of other types of low molecular weight materials also have been detected in SLE sera by this method. Approximately 75% of hypocomplementemic SLE sera tested had C1q reactants which in the majority of cases could be characterized as high or low molecular weight material, while sera from inactive patients were uniformly negative.

The exact nature of these reactants is at present unknown. Materials other than complexes of γ-globulin can react with C1q in gel diffusion; these include biologic polyanions such as NDNA, SDNA, double-stranded and single-stranded polyribonucleotides, heparin and endotoxin lipopolysaccharide, but these ordinarily can be differentiated from

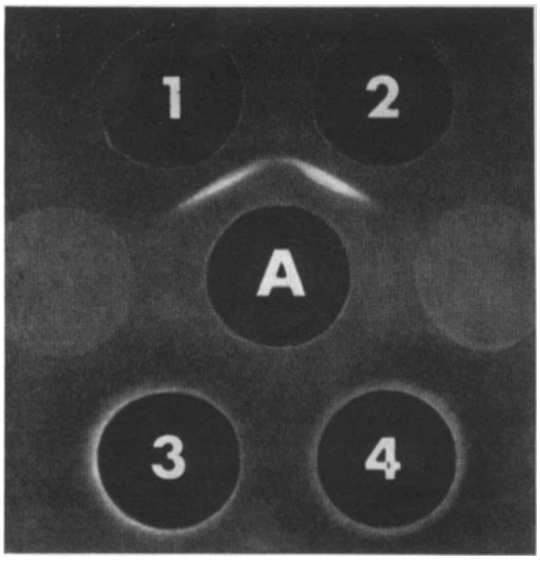

Fig. 5. *Gel diffusion plate showing the precipitation of isolated C1q (well A) with two sera from patients with active SLE disease and hypocomplementemia (wells 1 and 2).* The sera from two patients with inactive SLE are in wells 3 and 4.

γ-globulin reactants by their persistence after reduction and alkylation—a treatment which destroys the precipitability of γ-globulin complexes with C1q [40–44]. The high molecular weight reactants are comprised mainly of γG-globulins, although unidentified material is also present which could possibly represent antigens of immune complexes [41].

The low molecular weight complexes are also incompletely characterized. In isolated preparations of these reactants γG-globulin is present [41]. However, recent experiments indicate that material resistant to reduction and alkylation appears to be involved in some cases. Also, pronase treatment of the serum occasionally results in a C1q reactant of smaller size. These findings raise the possibility that a low molecular weight anionic substance, possibly of foreign origin, may be involved directly or complexed to γ-globulin.

The γ-globulin–anti-γ-globulin system. The correlation of the occurrence of serum cryoglobulin precipitates with periods of acute disease activity and especially nephritis has been pointed out by Christian et al [45, 46]. In the course of studies on C1q reactants in SLE sera it became apparent that in some cases a relationship exists between the presence of C1q precipitable complexes and cryoprecipitates [41]. The course of one patient whose episodes of disease activity correlated with the occurrence of C1q precipitins and cryoprecipitates in the serum is shown in Fig. 6. Two episodes of disease activity are shown. During these periods C1q precipitins and cryoprecipitates were present in the serum, associated with low serum complement and the initiation of a nephritic process.

Isolation work on these precipitates has offered evidence for unknown antigen-antibody complexes which interact

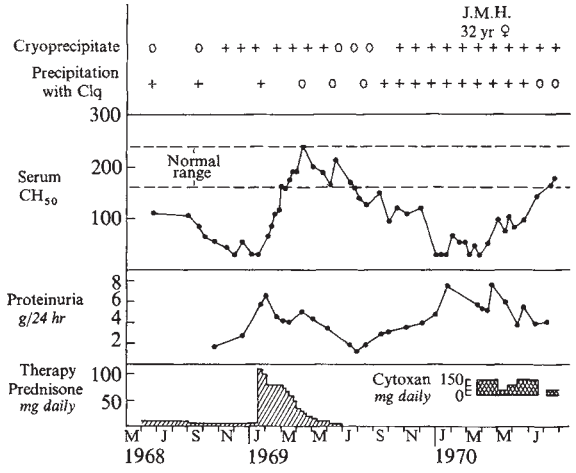

Fig. 6. *Serial study of an exceptional SLE patient (J. M. H.) showing two broad periods of clinical activity with renal disease which were associated with the presence of cryoprecipitins and C1q reactivity in the serum.* No antibodies to polynucleotides were present.

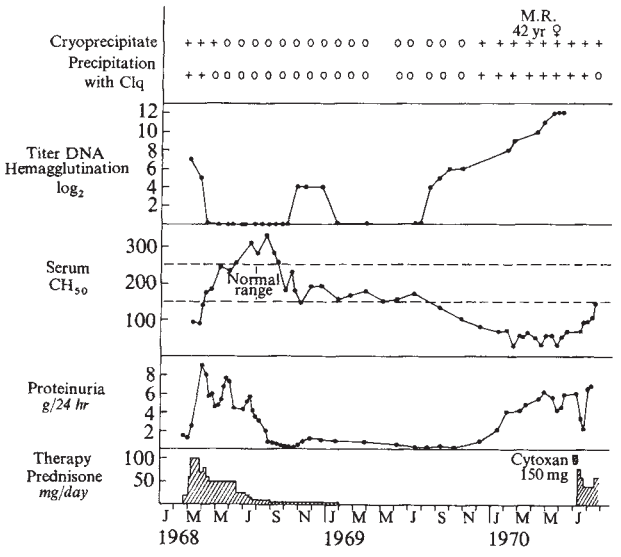

Fig 7. *Serial studies on patient M. R.* There are two periods of clinical exacerbations with nephritis that were associated with C1q reactants, cryoprecipitates and depressed serum complement. DNA antibodies were also present during these periods [36].

with IgM anti-γ-globulins and C1q to form larger complexes which appear to be of special significance. The complexes that appear to be involved in this system are similar to the high molecular weight type C1q reactants. Low molecular weight C1q reactants present in SLE are not reactive with anti-γ-globulins, and while they are associated with hypocomplementemia and clinical activity, they have not thus far been implicated in the nephritic process [41].

Further evidence for the possible significance of the γ-globulin–anti-γ-globulin system in the nephritic process was obtained by serologic and renal biopsy studies on a group of patients followed serially (Table 6). The first four all had relatively large amounts of cryoprecipitates which contained anti-γ-globulins. These patients had significant proteinuria and on renal biopsy showed heavy deposits of γM in a granular pattern in their glomeruli. In three of the biopsies anti-γ-globulins were detected by staining with fluorescein-labeled aggregated γ-globulin. The last three had smaller amounts of cryoprecipitates and no detectable anti-γ-globulins. These patients did not have significant proteinuria or γM deposits. The deposits that were present were of the more benign mesangial type.

Cryoglobulinemia in this situation appears to be an *in vitro* manifestation of circulating complexes which *in vivo* are deposited in the glomeruli. More direct evidence for this was obtained by demonstrating that the same anti-γ-globulins were in the isolated cryoprecipitins and glomerular deposits. This was accomplished by staining the renal deposits with fluorescein-labeled idiotypic antiserum to the isolated anti-γ-globulins [41, 54].

Table 6. Serological and immunofluorescent findings in SLE patients with cryoprecipitins and C1q precipitin reactions

	Cryoprecipitins			Renal biopsy				
	mg/100 ml	Rheumatoid factor	C1q precipitation	Pattern of deposit	γG	γM	RF	Proteinuria *g/24 hr*
M. R.	36.8	+	++	Granular	3+	3+	2+	4.8
J. M. H.	14.4	+	+++	Granular	3+	2+	Tr	5.7
J. H.	10.0	+	++	Granular	3+	3+	2+	1.4
A. B.[a]	12.0	+	++	Granular	1+	3+	0	1.6
G. G.	2.8	0	++	Mesangial	2+	Tr	NS[b]	0
S. V.	3.6	0	++	Mesangial	3+	0	NS	0.1
D. S.	2.7	0	+	Mesangial	3+	0	NS	0

[a] with Sjögren's syndrome.
[b] NS—not studied.

This system appears to be independent of the DNA-antiDNA system. It was noted in the studies on polynucleotide antibodies that three of twenty-five patients did not develop such antibodies during typical clinical courses of SLE, although renal disease was observed in two of the three patients. The patient whose course is shown in Fig. 6 had no evidence of antipolynucleotide antibodies. In addition, one other (A. B.) of the four patients (Table 6) with similar clinical and serologic findings to those shown in Fig. 6 did not have any detectable DNA antibodies. In another (Fig. 7), high-titered DNA antibodies occurred during periods of disease activity. These antibodies did not appear, however, to be related to the cryoprecipitins or C1q precipitins also present, since they were not concentrated in either of these precipitates and DNAse treatment of the serum did not affect formation of the cryoprecipitins or serum reactivity of C1q.

Discussion

The factors which determine the localization of complexes to the glomerular basement membrane (GBM) are incompletely understood. One possibility that arises from the above studies is that there is a progressive renal deposition of immunoglobulins and complement in the nephritis of SLE which may be correlated closely with the clinical course of the disease. The mesangium may be the initial site of localization of complexes followed by granular deposits along the basement membrane, and then, with advanced nephritis, a coalescence of these deposits may occur with formation of lumpy aggregates. There is some support for this thesis. The mesangium is known to contain phagocytic cells that represent a primary filtering agent of the kidney [47]. It is conceivable then that deposition of immune complexes on the GBM occurs only after saturation of the mesangium. Also, the lumpy pattern found in patients with more advanced nephritis is the one most frequently observed in nephritic kidneys at necropsy [48]. On the other hand in the present studies there was little correlation between the length of systemic disease and the type of deposit in the renal biopsy studies.

The finding of occasional renal biopsies that show granular deposits but very few deposits in the mesangium raises an alternative possibility that a qualitatively different type of immune complex with strong affinity for the GBM may be formed at certain stages of the disease and localize primarily on the GBM without entering the mesangium. From experimental evidence it is known that the size of the complexes as determined by ratio of antigen to antibody, and the presence of multivalent antigens, may influence the site of glomerular localization of complexes [8, 49–51]. Another consideration may be the ability of the antigen to activate complement. Certain biological antigens including many polynucleotides are known to activate complement [40–44]. The presence of both complement-activating antigen and antibody may enhance the nephrotoxicity of such complexes.

Linear localization of γG-globulin in glomeruli in SLE appears to be a special situation and was observed in two cases. The pattern of staining observed is analogous to that observed for anti-basement membrane antibodies in Goodpasture's syndrome but without concurrent localization of complement and with no histological or clinical evidence of glomerular damage. Also, there appears to be no sensitization of the basement membrane for localization of anti-γ-globulins, since both patients had high-titer rheumatoid factors at various times. On the contrary, the possibility exists that the γG-globulin in these cases represents non-nephrotoxic antibody because of either its inability to fix complement, or more likely, the spacing of its specific antigen on the GBM. The net effect then may be a protective one.

One of the factors which is difficult to assess is the effect of steroid therapy on glomerular deposits. From the data presented here there was no apparent relationship between therapy and pattern of staining. Experimental evidence, however, indicates that steroids can prevent deposition of immune complexes on the GBM and clinical nephritis in the acute serum sickness model but only partially affect the nephritis in a variable way in the chronic serum sickness model [52]. Since in man SLE appears to have acute and chronic periods of disease activity, the effect of steroids on the deposition of complexes may depend on when they are administered in the course of the disease.

A wide variety of antibodies to autologous antigens occur in the serum of patients with SLE. The antibodies which assume importance in relation to renal lesions are those which may come in contact with antigens in the bloodstream and form antigen-antibody complexes. Thus far NDNA and SDNA have been the only antigens demonstrated to occur in SLE sera in significant concentrations. Undoubtedly others occur. Elution studies of isolated glomeruli show concentrations of antiribonucleoprotein antibodies in addition to NDNA and SDNA antibodies. Antibodies to DSRNA, however, were not present. From these studies, one may expect the occurrence of ribonucleoprotein in the circulation.

A subject of considerable current interest is the source of NDNA and SDNA in SLE sera. The finding of uniquely high concentrations of SDNA appears particularly significant. Most antiNDNA antibodies can also react with SDNA [18], and SDNA-antiNDNA complexes have been shown to occur in addition to the NDNA-antiNDNA and SDNA-antiSDNA complexes. In contrast to NDNA, which occurs in high concentrations in other disease states, the highest concentrations of SDNA occur in SLE. Since high-titered NDNA antibodies are unique to SLE, while SDNA antibodies are not, the SDNA-antiNDNA system may be of considerable importance. It remains to be determined if the DNA present in renal lesions is predominantly native or single-stranded.

The high concentrations of SDNA present in SLE sera suggest that these antigens are mainly of host origin. It is possible, however, that a portion of these are of exogenous origin. The characterization of specificities of antibodies present has not so far determined the source of circulating antigens, since DNA antibodies are found to react similarly with DNA from different species, lower organisms and viruses [18]. Further studies are needed to resolve this point.

The "γ-globulin–anti-γ-globulin" system which is independent of the DNA systems appears to involve undetermined types of immune complexes that are detected in the circulation by precipitation with C1q. *In vivo* these complexes can interact with anti-γ-globulins and complement and deposit in tissues; *in vitro* this interaction is manifested by "cryoglobulinemia." Evidence that this system may play a role in the nephritic process is provided by studies which demonstrate the same anti-γ-globulins in the patient's cryoprecipitate and glomerular deposits by use of fluorescein-labeled antibody with idiotypic specificity for the anti-γ-globulins.

A large percentage of SLE patients have circulating rheumatoid factors, and not all of these have nephritis. It appears that circulating anti-γ-globulins have a detrimental effect in SLE patients only when circulating high molecular weight complexes such as those precipitable by C1q are also present. It remains possible, however, that specific characteristics of the anti-γ-globulins such as the affinity coefficient or thermal reactivity may also play a primary role. It is notable that in the patient in this study with the most severe nephritis, the anti-γ-globulins were of the cold reactive type.

Several types of immune complexes appear to be involved in the nephritis of SLE, and multiple types can occur in the individual patient. From the accumulated evidence the DNA-antiDNA complexes appear to be of major importance. However, the presence of these complexes does not invariably result in clinical nephritis. Also, DNA antibodies sometimes occur during periods of disease activity along with C1q reactants and cryoprecipitins. At this point, it cannot be determined which of these is the primary system involved in the nephritis in such cases. The stimulus for the formation of various antibodies and release of antigen in SLE is not known. Recent studies have suggested viral infection [55–59]; however, the evidence for this is sparse. It is of interest that in NZB/NZW mice, nephritis can be enhanced by various viral infections, but regardless of the virus used, the antinuclear antibodies comprised the major portion of the γ-globulin eluted from the affected kidneys of these animals, with specific antiviral antibodies making up a significant but lesser amount [60]. The situation may be similar in human SLE. Immune complexes involving polynucleotides may very well play the major role in the renal disease. However, unknown antigens, some of them perhaps detected as the unidentified C1q reactants, may be of special etiologic significance.

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